

Set	Items	Description
S1	10553	IPP
S2	950	ISOPENT? (W) PYROPHOSPHATE?
S3	161	ISOPENT? (W) PYROPHOSPHATE? (W) ISOMERASE?
S4	88774	CAROTENE? OR CAROTENOID?
S5	16	S1 AND S3
S6	39	S3 AND S4
S7	52	S5 OR S6
S8	25	RD (unique items)

?t s8/3,ab/all

>>>No matching display code(s) found in file(s): 43, 129-130, 140, 158, 173, 187, 189, 376, 428-429, 441, 446, 449, 452-453, 455-456, 636

8/3,AB/1 (Item 1 from file: 155)

DIALOG(R)File 155:MEDLINE(R)

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09563158 98256022

Expression of an active phytoene synthase from Erwinia uredovora and biochemical properties of the enzyme.

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Biochim Biophys Acta (NETHERLANDS) May 20 1998, 1392 (1) p51-8, ISSN 0006-3002 Journal Code: AOW

Languages: ENGLISH

Document type: JOURNAL ARTICLE

The crtB gene encoding phytoene synthase from the carotenogenic enterobacterium Erwinia uredovora was overexpressed to about 20% of the total cellular protein in Escherichia coli. Formation of the active phytoene synthase had the effect of suppressing the growth of the expressing strain. Presumably inhibition of growth arose from the depletion of the substrate geranylgeranyl pyrophosphate (GGPP) which, in E. coli, is necessary for the synthesis of essential prenylpyrophosphate derivatives. In order to overcome the poor growth characteristics of the phytoene synthase expressing strain, GGPP levels were increased by co-expressing the isoprenoid biosynthetic genes crtE and idi, encoding the Erwinia GGPP synthase and Rhodobacter isopentenyl pyrophosphate isomerase, respectively. The crude enzyme preparation was partially purified 15-fold by chromatography on a DEAE column. A non-radioactive assay was developed that enabled the conversion of GGPP to phytoene. The reaction product was identified by co-chromatography with authentic standards on HPLC systems and comparison of spectral characteristics. The phytoene formed in vitro was present in both a 15-cis and all-trans isomeric configuration. The essential cofactors required were ATP in combinations with either Mn2+ or Mg2+. The Km value for GGPP was determined as 41 microM. Phytoene synthesis was inhibited by phosphate ions and squalenstatin. The I50 value for the latter inhibitor was 15 microM. Lineweaver-Burk plots showed constant Km values in the presence or absence of squalenstatin. Copyright 1998 Elsevier Science B.V. All rights reserved.

8/3,AB/2 (Item 1 from file: 5)

DIALOG(R)File 5:BIOSIS PREVIEWS(R)

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13680492 BIOSIS Number: 99680492

Differential expression of two isopentenyl pyrophosphate isomerases and enhanced carotenoid accumulation in a unicellular chlorophyte

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Plant Physiology (Rockville) 114 (3 SUPPL.). 1997. 187.

Full Journal Title: PLANT BIOLOGY '97: 1997 Annual Meetings of the American Society of Plant Physiologists and the Canadian Society of Plant Physiologists, Japanese Society of Plant Physiologists and the Australian Society of Plant Physiologists, Vancouver, British Columbia, Canada, August 2-6, 1997. Plant Physiology (Rockville)

ISSN: 0032-0889

Language: ENGLISH

Document Type: CONFERENCE PAPER

Print Number: Biological Abstracts/RRM Vol. 049 Iss. 009 Ref. 161159

8/3,AB/3 (Item 2 from file: 5)
DIALOG(R)File 5:BIOSIS PREVIEWS(R)
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11782899 BIOSIS Number: 98382899

Terpenoid indole alkaloid biosynthesis and enzyme activities in two cell lines of *Tabernaemontana divaricata*

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Phytochemistry (Oxford) 39 (2). 1995. 341-349.

Full Journal Title: Phytochemistry (Oxford)

ISSN: 0031-9422

Language: ENGLISH

Print Number: Biological Abstracts Vol. 100 Iss. 005 Ref. 074737

The possible limitation of the rate of biosynthesis of terpenoid indole alkaloids by low enzyme levels was investigated in two cell lines of *Tabernaemontana divaricata* with different terpenoid indole alkaloid biosynthetic capacities. The activities of tryptophan decarboxylase (TDC), strictosidine synthase (SSS), strictosidine glucosidase (SG), **isopentenyl pyrophosphate isomerase (IPP isomerase)** and geraniol 10-hydroxylase (G10H) of both cell lines were compared. The activities of TDC, SSS and **IPP isomerase** did not show a direct relationship with the biosynthetic capacity but SG and G10H might be limiting. In order to test whether the availability of the terpenoid precursor limits the biosynthesis of the terpenoid indole alkaloids, loganin was fed to the cultures. Loganin-feeding did not influence any of the measured enzyme activities but increased the terpenoid indole alkaloid accumulation of both cell lines to similar levels. A five-fold increase was observed for the accumulating line and a more than 100-fold increase for the low-accumulating one. Strictosidine accumulated mainly in the low-accumulating cell line which has high TDC and low SG activity; the amounts and types of the other terpenoid indole alkaloids which accumulated were similar in both lines. From this it can be concluded that the biosynthesis of terpenoid indole alkaloids in both cultures is limited by the availability of terpenoid precursors; this pathway is not saturated with substrates under normal culture conditions.

8/3,AB/4 (Item 3 from file: 5)
DIALOG(R)File 5:BIOSIS PREVIEWS(R)
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11483339 BIOSIS Number: 98083339

Effects of clomazone on IPP isomerase and prenyl transferase activities in cell suspension cultures and cotyledons of solanaceous species

Scott J E; Weston L A; Chappell J; Hanley K

Dep. Horticulture and Landscape Arch., Univ. Kentucky, Lexington, KY 40546, USA

Weed Science 42 (4). 1994. 509-516.

Full Journal Title: Weed Science

ISSN: 0043-1745

Language: ENGLISH

Print Number: Biological Abstracts Vol. 099 Iss. 004 Ref. 053749

Laboratory assays were conducted to determine the sensitivity of tomato and tobacco cell suspension cultures and tomato and pepper cotyledons to clomazone. A comparison of fresh weight and **carotenoid** content indicated up to a three-fold difference between the clomazone-tolerant tobacco and clomazone-susceptible tomato cell suspension cultures. In contrast, an approximate 60-fold difference between the tolerant pepper and susceptible tomato cotyledons was observed when total chlorophyll and **carotenoid** contents were measured. The effect of clomazone and its possible metabolites on in vivo and in vitro extractable **IPP isomerase (EC**

5.3.3.2) and prenyl transferase (EC 2.5.1.29) activity was investigated. There was no clear inhibitory effect of clomazone or possible clomazone metabolites upon enzyme activity in tomato or tobacco cell suspension cultures or on light or dark grown tomato or pepper cotyledons. No specific enzymatic target site of clomazone was identified in correlation with the reduction in total chlorophyll or **carotenoid** content.

8/3,AB/5 (Item 4 from file: 5)
DIALOG(R)File 5:BIOSIS PREVIEWS(R)
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11228731 BIOSIS Number: 97428731

Plant phytoene synthase complex: Component enzymes, immunology, and biogenesis

Camara B

Inst. Biol. Mol. Plantes, Cent. Natl. Rech. Sci., F-67084 Strasbourg, FRA
0 (0). 1993. 352-365.

Full Journal Title: Packer, L. (Ed.). Methods in Enzymology, Vol. 214. Carotenoids, Part B: Metabolism, genetics, and biosynthesis. xxvii+468p. Academic Press, Inc.: San Diego, California, USA; London, England, UK. ISBN 0-12-182115-3.

ISSN: 0076-6879

Language: ENGLISH

Document Type: BOOK

Print Number: Biological Abstracts/RRM Vol. 046 Iss. 010 Ref. 153495

8/3,AB/6 (Item 5 from file: 5)
DIALOG(R)File 5:BIOSIS PREVIEWS(R)
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11177595 BIOSIS Number: 97377595

Light-stimulated carotenoid biosynthesis during transformation of maize etioplasts is regulated by increased activity of isopentenyl pyrophosphate isomerase

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Plant Physiology (Rockville) 105 (2). 1994. 529-534.

Full Journal Title: Plant Physiology (Rockville)

ISSN: 0032-0889

Language: ENGLISH

Print Number: Biological Abstracts Vol. 098 Iss. 005 Ref. 066084

Light-stimulated **carotenoid** biosynthesis associated with the transformation of etioplasts to chloroplasts was investigated after dark-grown maize (*Zea mays*) seedlings were transferred into light. These studies focused on the enzymes of the pathway to detect those enzyme activities that were stimulated in the light and thus that were responsible for increased biosynthesis of **carotenoids**. In preliminary experiments, norflurazon, an inhibitor of phytoene desaturase, was used to prevent phytoene being further metabolized to **carotenoids**. Light-dependent stimulation of phytoene accumulation indicated that the light-regulated steps are located in the pathway leading to phytoene synthesis. The use of the ¹⁴C-labeled precursors mevalonic acid, isopentenyl pyrophosphate, and farnesyl pyrophosphate pointed to increased activity of an enzyme involved in the biosynthetic steps between isopentenyl pyrophosphate and farnesyl pyrophosphate. Determination of the activities of all five enzymes of the pathway involved in the sequence from mevalonic acid to phytoene revealed that the only enzyme activity stimulated by light was **isopentenyl pyrophosphate isomerase**. Over a 3-h period of illumination, this enzyme activity, like **carotenoid** biosynthesis, was stimulated 2.8-fold.

8/3,AB/7 (Item 6 from file: 5)
DIALOG(R)File 5:BIOSIS PREVIEWS(R)
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9527721 BIOSIS Number: 94032721

CLOMAZONE DOES NOT INHIBIT THE CONVERSION OF ISOPENTENYL PYROPHOSPHATE TO GERANYL FARNESYL OR GERANYLGERANYL PYROPHOSPHATE IN-VITRO

CROTEAU R

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PLANT PHYSIOL (BETHESDA) 98 (4). 1992. 1515-1517. CODEN: PLPHA

Full Journal Title: Plant Physiology (Bethesda)

Language: ENGLISH

Clomazone, an herbicide that reduces the levels of leaf **carotenoids** and chlorophylls, is thought to act by inhibiting **isopentenyl pyrophosphate isomerase** or the prenyltransferases responsible for the synthesis of geranylgeranyl pyrophosphate. Cell-free extracts prepared from the oil glands of common sage (*Salvia officinalis*) are capable of converting isopentenyl pyrophosphate to geranylgeranyl pyrophosphate. Clomazone at 250 micromolar (a level that produced leaf bleaching) had no detectable effect on the activity of the relevant enzymes (**isopentenyl pyrophosphate isomerase** and the three prenyltransferases, geranyl, farnesyl, and geranylgeranyl pyrophosphate synthases). Thus, inhibition of geranylgeranyl pyrophosphate biosynthesis does not appear to be the mode of action of this herbicide.

8/3,AB/8 (Item 7 from file: 5)

DIALOG(R)File 5:BIOSIS PREVIEWS(R)

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9131170 BIOSIS Number: 93116170

HERBICIDE CLOMAZONE DOES NOT INHIBIT IN-VITRO GERANYLGERANYL SYNTHESIS FROM MEVALONATE

WEIMER M R; BALKE N E; BUHLER D D

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PLANT PHYSIOL (BETHESDA) 98 (2). 1992. 427-432. CODEN: PLPHA

Full Journal Title: Plant Physiology (Bethesda)

Language: ENGLISH

Clomazone reduced the chlorophyll and **carotenoid** contents of spinach (*Spinacia oleracea* L.), barley (*Hordeum vulgare* L.), velvetleaf (*Abutilon theophrasti* Medik.), and soybean (*Glycine max* L. Merr.) seedlings. The order of species sensitivity was velvetleaf > spinach > barley > soybean. Clomazone (100 micromolar) did not affect the in vitro activities of spinach **isopentenyl pyrophosphate isomerase** or prenyl transferase. Clomazone also did not affect the synthesis of isopentenyl pyrophosphate from mevalonic acid. Thus, clomazone had no direct in vitro effect on the synthesis of geranylgeranyl pyrophosphate from mevalonic acid. Greening seedlings of both soybean and velvetleaf metabolized clomazone. No qualitative differences in the metabolites were detected between soybean and velvetleaf. Thus, differential metabolism of clomazone to a toxic chemical that inhibits terpenoid synthesis is unlikely. Clomazone has either a mode of action not yet identified or a metabolite that is selective in that it is much more active in sensitive than tolerant species.

8/3,AB/9 (Item 8 from file: 5)

DIALOG(R)File 5:BIOSIS PREVIEWS(R)

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8326580 BIOSIS Number: 41010580

CAROTENOID BIOSYNTHESIS AND REGULATION IN PLANTS

WALTER J; HUGUENEY P; D'HARLINGUE A; LAUNAY J; CAMARA B

BIOCHIMIE REGULATIONS CELLULAIRES, UNIV. BORDEAUX I, AVE. DES FACULTES, 33405 TALENCE CEDEX, FR.

201ST ACS NATIONAL MEETING OF THE AMERICAN CHEMICAL SOCIETY, ATLANTA, GEORGIA, USA, APRIL 14-19, 1991. ABSTR PAP AM CHEM SOC 201 (1-2). 1991.

AGFD 71. CODEN: ACSRA

Language: ENGLISH

Document Type: CONFERENCE PAPER

8/3,AB/10 (Item 9 from file: 5)
DIALOG(R)File 5:BIOSIS PREVIEWS(R)
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8110906 BIOSIS Number: 91031906

**SITE OF CLOMAZONE ACTION IN TOLERANT SOYBEAN AND SUSCEPTIBLE COTTON
PHOTOMIXOTROPHIC CELL SUSPENSION CULTURES**

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PLANT PHYSIOL (BETHESDA) 94 (2). 1990. 704-709. CODEN: PLPHA

Full Journal Title: Plant Physiology (Bethesda)

Language: ENGLISH

Studies were conducted to determine the herbicidal site of clomazone action in tolerant-soybean (*Glycine max* [L.] Merr. cv Corsoy (SB-M) and susceptible-cotton (*Gossypium hirsutum* [L.] cv Stoneville 825) (COT-M) photomixotrophic cell suspension cultures. Although a 10 micromolar clomazone treatment did not significantly reduce the terpene or mixed terpenoid content (microgram per gram fresh weight) of the SB-M cell line, there was over a 70% reduction in the chlorophyll (Chl), carotenoid (CAR), and plastoquinone (PQ) content of the COT-M cell line. The tocopherol (TOC) content was reduced only 35.6%. Reductions in the levels of Chl, CAR, TOC, and PQ indicate that the site of clomazone action in COT-M cells is prior to geranylgeranyl pyrophosphate (GGPP). The clomazone treatment did not significantly reduce the flow of [¹⁴C]mevalonate ([¹⁴C]MEV) (nanocuries per gram fresh weight) into CAR and the three mixed terpenoid compounds of SB-M cells. Conversely, [¹⁴C]MEV incorporation into CAR and the terpene moieties of Chl, PQ, and TOC in COT-M cells was reduced at least 73%, indicating that the site of clomazone action must be after MeV. Sequestration of clomazone away from the chloroplast cannot account for soybean tolerance to clomazone since chloroplasts isolated from both cell lines incubated with [¹⁴C] clomazone contained a similar amount of radioactivity (disintegrations per minute per microgram of Chl). The possible site(s) of clomazone inhibition include mevalonate kinase, phosphomevalonate kinase, pyrophosphomevalonate decarboxylase, isopentenyl pyrophosphate isomerase, and/or a prenyl transferase.

8/3,AB/11 (Item 10 from file: 5)
DIALOG(R)File 5:BIOSIS PREVIEWS(R)
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5539544 BIOSIS Number: 32061851

**METABOLISM OF PLASTID TERPENOIDS IN-VITRO INHIBITION OF PHYTOENE
SYNTHESIS BY PHENETHYL PYROPHOSPHATE DERIVATIVES**

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FRANCE.

FEBS (FED EUR BIOCHEM SOC) LETT 210 (2). 1987. 211-215. CODEN: FEBLA

Full Journal Title: FEBS (Federation of European Biochemical Societies)
Letters

Language: ENGLISH

8/3,AB/12 (Item 11 from file: 5)
DIALOG(R)File 5:BIOSIS PREVIEWS(R)
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5421993 BIOSIS Number: 82066796

**ISOPRENOID SYNTHESIS IN ESCHERICHIA-COLI SEPARATION AND PARTIAL
PURIFICATION OF FOUR ENZYMES INVOLVED IN THE SYNTHESIS**

FUJISAKI S; NISHINO T; KATSUKI H

DEP. CHEM., FAC. SCI., KYOTO UNIV., SAKYO, KYOTO, KYOTO 606, JPN.

J BIOCHEM (TOKYO) 99 (5). 1986. 1327-1338. CODEN: JOBIA

Full Journal Title: Journal of Biochemistry (Tokyo)

Language: ENGLISH

Isopentenyl pyrophosphate (IPP) isomerase, farnesyl pyrophosphate (FPP)

synthetase, octaprenyl pyrophosphate (OPP) synthetase and undecaprenyl pyrophosphate (UPP) synthetase were partially purified from *Escherichia coli* by DEAE-Toyopearl chromatography. FPP synthetase catalyzed the condensation of IPP with dimethylallyl pyrophosphate (DPP) as well as with geranyl pyrophosphate (GPP) to yield FPP as final product. OPP synthetase and UPP synthetase catalyzed the condensation of IPP with FPP to yield OPP and cis,trans-polyprenyl pyrophosphates (the C45-, C50-, and C55-compound), respectively. Neither DPP nor GPP acted as a priming substrate for either enzyme. These four enzymes required Mg²⁺ or Mn²⁺ for their activities. UPP synthetase required also Triton X-100 for its activity. The addition of Triton X-100 enhanced OPP synthetase, but it did not affect IPP isomerase and FPP synthetase. It seems possible that the combination of the four enzymes ensures the *in vivo* synthesis of long-chain isoprenoids in *E. coli*.

8/3,AB/13 (Item 12 from file: 5)
DIALOG(R)File 5:BIOSIS PREVIEWS(R)
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4133630 BIOSIS Number: 76083481

**BIOSYNTHESIS OF ALLYLIC ISOPRENOID PYRO PHOSPHATES BY AN ENZYME
PREPARATION FROM THE FLAVEDO OF CITRUS-PARADISI**

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PHYTOCHEMISTRY (OXF) 22 (2). 1983. 431-434. CODEN: PYTCA

Full Journal Title: PHYTOCHEMISTRY (Oxford)

Language: ENGLISH

A cell-free system obtained from *C. paradisi* flavedo transformed mevalonic acid into mono- and sesquiterpenoids of E- and Z-conformation. The enzyme system required bivalent metal ions and the presence of SH groups. IPP [isopentenyl pyrophosphate] isomerase activity (EC 5.3.3.2) was independent of metal ions and almost insensitive to SH group reagents, while prenyltransferase (EC 2.5.1.1) was inactivated by DTNB and required bivalent metals for activity. The nature of the metal ion defined the stereochemistry of the products formed by prenyltransferase. The ratio of E-Z farnesylpyrophosphates was 3:1. This *Citrus* sp. could be a good starting material for the study of the stereochemistry of the enzymes forming E and Z sesquiterpenoids.

8/3,AB/14 (Item 13 from file: 5)
DIALOG(R)File 5:BIOSIS PREVIEWS(R)
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2196152 BIOSIS Number: 64023072

**THE DIVISION OF DI METHYLALLYL PYRO PHOSPHATE FROM POLY ISO PRENOID TO
CYCLOPIAZONIC-ACID BIOSYNTHESIS IN PENICILLIUM-CYCLOPIUM**

MCGRATH R M; NOURSE P N; NEETHLING D C; FERREIRA N P

BIOORG CHEM 6 (1). 1977 53-69. CODEN: BOCMB

Full Journal Title: Bioorganic Chemistry

During the production of .alpha.-cyclopiazonic acid (.alpha.CA) by *P. cyclopium*, dimethylallyltransferase (EC 2.5.1.1) T, isopentenyl pyrophosphate isomerase (EC 5.3.3.2) I, and a prenyl-aryltransferase, S, which produces .beta.-cyclopiazonic acid (.beta.CA) are induced at the same time. This last enzyme appears maximally before the highest rate of .alpha.- or .beta.CA production. Both transferases are not utilized to their maximum capacity, and the production of their end products seems to bear no relationship to their concentrations. Other controls must play an important role in the utilization of their common substrate dimethylallylpyrophosphate (DMAPP). There are 2 possible control systems; a direct competition by S and T for DMAPP and control by compartmentation. The 1st possibility is the more likely, in view of some of the controls that could apply to the deflection. The 3 enzymes were separated so that possible controls on the deflection of DMAPP from polyisoprenoids could be studied. They possessed a subunit structure and exhibited maximum MW (in the absence of divalent cations and presence of a thiol reductant) of

96,000 (S) and 64,000 (I and T) daltons. Mg²⁺ caused a diminution in size to 75,000 (S) and 50,000 (I and T) daltons. Mn²⁺ had the same effect on I and T but caused major disruptive changes to S. These effects were reversible by addition of EDTA. S was specific for DMAPP and cycloacetoacetyl-L-tryptophan (cAATrp) and exhibited Km as follows; **GRAPHIC**. 6.0 .mu.M and **GRAPHIC**. 2.0 .mu.M. It had no obvious requirement for a divalent cation and had an isoelectric point of 5.3. I had a Km of 6.7 .mu.M and an isoelectric point of 4.5, and either Mg²⁺ or Mn²⁺ was essential. The Km for T could not be given but its isoelectric point was 5.1. The enzyme carried out the 2 reactions normally associated with it (i.e., 2 additions of IPP to produced farnesyl pyrophosphate) and required Mg²⁺ to do so. The pH optima for S, I, and T were 6.5-7.5, 6.0 and 8.0, respectively. The early controlling factor was the appearance of the consubstrate of S, cAATrp. Other factors were the appearance of .alpha.CA which inhibited T more effectively than S, the removal of free Mn²⁺ and Mg²⁺, both essential for I and T but not for S, possibly brought about by chelation with cAATrp, .alpha.- and .beta.CA, the observed low pH of 6.0 when the activity of S was unaltered, I was at its highest, and T exhibited 50% of its maximum and an activation of I by low physiological levels of .beta.CA and cAATrp which would enhance the rate of appearance of DMAPP to react with an existing pool of cAATrp.

8/3,AB/15 (Item 1 from file: 34)

DIALOG(R)File 34:SciSearch(R) Cited Ref Sci
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04254421 Genuine Article#: RT069 Number of References: 22

Title: UTILIZATION OF PHOTOSYNTHATES DURING RUBBER BIOSYNTHESIS IN GUAYULE (PARTHENIUM-ARGENTATUM GRAY)

Author(s): REDDY AR; DAS VSR

Corporate Source: PONDICHERRY UNIV,SCH LIFE SCI/PONDICHERRY 605011//INDIA//;
PONDICHERRY UNIV,SCH LIFE SCI/PONDICHERRY 605011//INDIA//; UNIV
HYDERABAD,SCH LIFE SCI/HYDERABAD 500134/ANDHRA PRADESH/INDIA/

Journal: PHOTOSYNTHETICA, 1995, V31, N3, P351-358

ISSN: 0300-3604

Language: ENGLISH Document Type: ARTICLE

Abstract: Incorporation of C-14 labelled CO₂, 3-phosphoglycerate (PGA), phosphoenolpyruvate (PEP) and pyruvate into hexane extractable rubber fractions in the cut shoots of guayule (Parthenium argentatum Gray) was determined in order to evaluate the role of photosynthesis in providing precursors for rubber biosynthesis. DCMU inhibited the incorporation of labelled CO₂ and PGA into rubber. The incorporation of (CO₂)-C-14 into rubber depended on irradiance. Enzymatic activities of phosphoglyceromutase, enolase, pyruvate kinase and pyruvate dehydrogenase complex found in purified chloroplasts from the leaves indicated the chloroplast autonomy for intraplastid acetyl coenzyme A formation. The enzymes related to the biosynthesis of isopentenyl pyrophosphate (IPP) were associated with both leaf and stem extracts. Rubber producing enzyme activities, namely IPP isomerase and rubber transferase, were abundantly localized in roots and stems of guayule while the leaves exhibited low activities of these enzymes. Hence the leaves of guayule play a major role in providing precursors for rubber formation in stems and roots.

8/3,AB/16 (Item 2 from file: 34)

DIALOG(R)File 34:SciSearch(R) Cited Ref Sci
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01948002 Genuine Article#: JN913 Number of References: 47

Title: LOCALIZATION OF THE ENZYME GERANYLGERANYLPYROPHOSPHATE SYNTHASE IN CAPSICUM FRUITS BY IMMUNOGOLD CYTOCHEMISTRY AFTER CONVENTIONAL CHEMICAL FIXATION OR QUICK-FREEZING FOLLOWED BY FREEZE-SUBSTITUTION - LABELING EVOLUTION DURING FRUIT RIPENING

Author(s): CHENICLET C; RAFIA F; SAINTGUILY A; VERNA A; CARDE JP

Corporate Source: UNIV BORDEAUX 1,PHYSIOL CELLULAIRE VEGETALE
LAB,CNRS,UNITE ASSOCIEE 568,AVE FAC/F-33405 TALENCE//FRANCE//; UNIV

BORDEAUX 2, NEUROCYTOCHIM CYTOL LAB, CNRS, UNITE ASSOCIEE 339/F-33405
TALENCE//FRANCE/

Journal: BIOLOGY OF THE CELL, 1992, V75, N2, P145-154

ISSN: 0248-4900

Language: ENGLISH Document Type: ARTICLE

Abstract: The enzyme geranylgeranylpyrophosphate synthase (GGPPS), which plays a key role in the synthesis of diterpene compounds, **carotenoids** and higher terpenoids, has been localized in Capsicum fruit cells by ultrastructural immunogold cytochemistry, after conventional chemical fixation of tissues and quick-freezing followed by freeze-substitution of isolated chloroplasts and chromoplasts. In agreement with previous biochemical studies on cell fractions, the enzyme seems restricted to the plastid compartment. Together with the phenotypic changes of the fruit and the ultrastructural modifications of the plastids during the transition of chloroplasts to chromoplasts, the amount of immunolabelling over plastid sections increases more than a ten-fold factor in the course of fruit ripening. In chemically fixed tissues, the gold labelling of chloroplasts is very faint and erratically localized whereas in further transition stages, and in chromoplasts, most of the gold particles surround the developing plastoglobuli, which are the characteristic **carotenoid**-bearing structures. Because of the very low and inconstant labelling of chloroplasts in green fruits after chemical fixation, cryofixed and acetone freeze-substituted purified plastids were used as a model system for an accurate localization of the enzyme in these organelles. Quick-freezing in buffered sucrose by slam-freezing on a cold copper block results in optimal preservation of the plastids and improved labelling of GGPPS. The enzyme is not scattered at random throughout the stroma. Gold particles are concentrated in distinct stroma regions, and especially at the sites of initiation of stroma globuli which are the early structural event of **carotenoid** accumulation. A few gold particles are also present on the margins of thylakoids and, presumably, on the plastid envelope. This paper reports further evidence of the central role of the plastid compartment in the production of C20 isoprenoid intermediates in the plant cell, shows the spatial relationship of the enzyme geranylgeranylpyrophosphate synthase with the plastid substructures and the existence of several GGPPS pools within the plastids. It demonstrates the interest of cryo-methods for an accurate localization of various enzymes in plant cells.

8/3,AB/17 (Item 3 from file: 34)

DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

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00767848 Genuine Article#: EV267 Number of References: 256

Title: THE BIOSYNTHESIS OF TRITERPENOID, STEROIDS, AND CAROTENOID

Author(s): HARRISON DM

Corporate Source: UNIV WARWICK, DEPT CHEM/COVENTRY CV4 7AL/W
MIDLANDS/ENGLAND/

Journal: NATURAL PRODUCT REPORTS, 1990, V7, N6, P459-484

Language: ENGLISH Document Type: REVIEW

8/3,AB/18 (Item 1 from file: 50)

DIALOG(R)File 50:CAB Abstracts

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02788360 CAB Accession Number: 931645813

Seasonal variations in rubber biosynthesis, 3-hydroxy-3-methylglutaryl-CoA reductase, and rubber transferase activities in Parthenium argentatum in the Chihuahuan Desert.

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Department of Biochemistry & Biophysics, Texas A&M University, College Station, TX 77843, USA.

Plant Physiology vol. 103 (2): p.535-542

Publication Year: 1993

ISSN: 0032-0889

Language: English

Document Type: Journal article

Rubber content and the activities of enzymes in the polyisoprenoid pathway of *P. argentatum* (guayule) were examined throughout the growing season in field plots in the Chihuahuan Desert, Texas, USA. Rubber content of the plants was low in July and August, then increased slowly, and from October to December there was a rapid increase in rubber formation (per plant) from 589.0 to 4438.0 mg. Percentage rubber in the plants increased from 0.7 (mg/g dry weight) in August and 1.27 in October to 5.5 in December. The rapid increase in rubber formation may have resulted from exposing the plants to low temperatures of 5 to 7 deg C. Activity of hydroxymethylglutaryl-CoA reductase (HMGR) was 21.1 nmol mevalonic acid (MVA)/h per g fresh weight in the bark of the lower stems in June during seedling growth, decreased to 5.1 nmol in July, 2.9 nmol in September, and from October to December the activity increased from 5.0 to 29.9 nmol. Activity of rubber transferase was 65.5 nmol isopentenyl pyrophosphate (IPP)/h per g fresh weight in the bark in September and increased to 357.5 nmol in December. The rapid increase in the activities of HMGR and rubber transferase coincided with the rapid increase in rubber formation. Activities of MVA kinase and IPP isomerase did not significantly increase in the autumn and winter. A tomato HMGR-1 cDNA probe containing a highly conserved C-terminal region of HMGR genes hybridized at low stringency with several bands on blots of HindIII-digested genomic DNA from guayule. In northern blots with the HMGR-1 cDNA probe at low stringency, HMGR mRNA was high in June and November, corresponding to periods of high HMGR activity during seedling growth and rapid increase in rubber formation. The seasonal variations in rubber formation and HMGR mRNA, HMGR activity, and rubber transferase activity may be due to low temperature stimulation in the autumn and winter months. 24 ref.

8/3,AB/19 (Item 2 from file: 50)

DIALOG(R)File 50:CAB Abstracts

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02301082 CAB Accession Number: 902300337

Interconversion of phenyl pyrophosphate and subsequent reactions in the presence of FMC 57020.

Sandmann, G.; Boger, P.

Lehrstuhl für Biochemie der Pflanzen, Universität, Postfach 5560, 7750 Konstanz, German Federal Republic.

Zeitschrift für Naturforschung. Section C, Biosciences vol. 42 (6): p.803-807

Publication Year: 1987

Language: English

Document Type: Conference paper; Journal article

This is the text of a paper given at an international workshop entitled 'Herbicides affecting chloroplast functions', held on 17-20 Aug. 1986, at Lake Placid, New York. In vitro studies of FMC 57020 (clomazone) on terpenoid biosynthesis showed that it acts at an early stage, affecting the conversion of isopentenyl pyrophosphate to geranylgeranyl pyrophosphate catalyzed by isopentenyl pyrophosphate isomerase and prenyl transferase. An inhibition of the carotenogenic enzymes phytoene desaturase, zeta - carotene desaturase, and lycopene cyclase could be excluded. Comparison of I50 values for in vivo chlorophyll, carotenoid, ergosterol and gibberellin biosynthesis as well as in vitro formation of phytoene, phytol and kaurene in various autotrophic and heterotrophic organisms showed that terpenoid biosynthesis in the chloroplast is much more strongly affected than extraplastidic terpenoid formation. 17 ref.

8/3,AB/20 (Item 1 from file: 73)

DIALOG(R)File 73:EMBASE

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10762033 EMBASE No: 98183119

Metabolic engineering for the production of carotenoids in non-carotenogenic bacteria and yeasts

Misawa N.; Shimada H.
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Journal of Biotechnology (Netherlands) , 1998, 59/3 (169-181)
CODEN: JBITD ISSN: 0168-1656
PUBLICATION DATE: 19980103
PUBLISHER ITEM IDENTIFIER: S0168165697001545
DOCUMENT TYPE: Journal Short Survey
LANGUAGES: ENGLISH SUMMARY LANGUAGES: ENGLISH
NUMBER OF REFERENCES: 50

The crt gene clusters responsible for the biosynthesis of **carotenoids** such as lycopene, beta-**carotene** and astaxanthin have been isolated from carotenogenic bacteria such as *Erwinia* species and the marine bacterium *Agrobacterium aurantiacum*. The functions of the individual genes have been identified. The first substrate of the enzymes encoded by the *Erwinia* crt clusters is farnesyl pyrophosphate which is not only the precursor for **carotenoid** biosynthesis but also sterols, dolichols and other numerous isoprenoid compounds. *Escherichia coli* does not naturally synthesize **carotenoids** , but by using the carotenogenic genes recombinant strains accumulating lycopene, beta-**carotene** and astaxanthin have been produced. Other non-carotenogenic bacteria such as *Zymomonas mobilis* have also been engineered to produce beta- **carotene** by the introduction of the corresponding crt genes. A gene capable of enhancing **carotenoid** levels in *E. coli* has also been isolated from cDNA libraries of the yeast *Phaffia rhodozyma* and the green alga *Haematococcus pluvialis*. This gene has been found to encode an **isopentenyl pyrophosphate isomerase** . It has further been shown that the edible yeasts *Candida utilis* as well as *Saccharomyces cerevisiae*, which possess no **carotenoid** biosynthetic pathway, acquire the ability to produce **carotenoids** , when the carotenogenic genes are expressed under the control of yeast-derived promoters and terminators. It has been observed in the yeasts *S. cerevisiae* and *C. utilis* carrying the lycopene biosynthesis genes that ergosterol content is decreased by 10 and 35%, respectively. It is therefore likely that the carbon flux for the ergosterol biosynthesis has been partially directed from farnesyl pyrophosphate to a new pathway for the lycopene biosynthesis. Further, the expression of a truncated gene which codes for the catalytic domain of the endogenous 3-hydroxy-3-methylglutaryl coenzyme A reductase, has been found to be effective for enhancing **carotenoid** levels in the yeast *C. utilis*.

8/3,AB/21 (Item 1 from file: 144)
DIALOG(R)File 144:Pascal
(c) 1998 INIST/CNRS. All rts. reserv.

10302182 PASCAL No.: 92-0250482
Localisation des activites IPP-isomerase et prenyltransferases dans les cellules de Vitis vinifera cv Muscat de Frontignan cultivees in vitro
(Localization of IPP-isomerase and phenyltransferase activities in cells of *Vitis vinifera* cv Muscat de Frontignan cultivated in vitro)
FERON Gilles; AMBID Christian, dir
Univ.: Institut national polytechnique Toulouse. FRA Degree: Th. doct.
: Biochim.

1991-05; 1991 89 p.
Language: French Summary Language: French; English
L' **isopentenyl pyrophosphate isomerase** et les prenyltransferases, enzymes clefs de la voie de biosynthese des terpenoides sont etudiees dans le but d'identifier les facteurs responsables de l'absence d'accumulation de monoterpenes chez des suspensions cellulaires de *Vitis vinifera* cv Muscat de Frontignan. Ces enzymes peuvent etre impliquees dans la voie de biosynthese des terpenoides mais aussi des steroides. La localisation subcellulaire de ces enzymes est realisee a partir de protoplastes de raisin Muscat obtenus par digestion enzymatique de la paroi cellulaire. Les resultats demontrent que les activites **IPP -isomerase** et prenyltransferases sont localisees surtout dans le cytosol, mais aussi dans une fraction membranaire obtenue apres purification d'un culot brut sur gradient de saccharose ou de Percoll. L'examen approfondi de cette fraction a l'aide d'enzymes marqueurs des mitochondries, des microbodies et des

plastest, revele une origine plastidiale. Des observations de cette fraction en microscopie electronique montrent que les plastest purifies sur gradient de Percoll sont intactes. De plus des incubations de la fraction soluble et de la fraction plastidiale en presence de (1- SUP 1 SUP 4 C)IPP demontrent la presence de deux activites prenyltransferases: une geranyl pyrophosphate synthetase dans le plaste et une farnesyl pyrophosphate synthetase dans le cytosol. La compartimentation specifique de ces enzymes peut etre une explication pour la perte de production de monoterpenes observee chez des cellules cultivees in vitro. Le probleme de la sequestration de la GPP synthetase au sein des plastest, lie a la permeabilite des organites pour les substrats de l'enzyme est aborde

8/3,AB/22 (Item 1 from file: 315)

DIALOG(R)File 315:ChemEng & Biotec Abs

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442075 CEABA Accession No.: 29-08-014100 DOCUMENT TYPE: Patent

Title: **Genes of carotenoid biosynthesis and metabolism and a system for screening for such genes.**

AUTHOR: Cunningham, F. X., Jr. ; Sun, Z.

CORPORATE SOURCE: Univ. Maryland College Park College Park, MD 20742 USA

CODEN: PIXXD2

PATENT NUMBER: WO 9736998

PUBLICATION DATE: 9 Oct 1997 (971009) LANGUAGE: English

PRIORITY PATENT APPLICATION(S) & DATE(S): US 8624125 (960329)

ABSTRACT: The DNA sequences for eukaryotic genes encoding cyclase, **isopentenyl pyrophosphate isomerase** and **.beta.- carotene hydroxylase** and vectors and hosts containing the genes are disclosed. Methods for controlling the ratio of various **carotenoids** in a host and the production of novel **carotenoid** pigments are also described.

8/3,AB/23 (Item 1 from file: 357)

DIALOG(R)File 357:Derwent Biotechnology Abs

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218242 DBA Accession No.: 97-13363 PATENT

Eukaryotic carotenoid biosynthetic enzymes and related genes-

epsilon-cyclase, isopentenyl- pyrophosphate- isomerase and beta-carotene-hydroxylase gene expression in e.g. Escherichia coli for carotenoid production

AUTHOR: Cunningham Jr F X; Sun Z

CORPORATE SOURCE: College Park, MD, USA.

PATENT ASSIGNEE: Univ.Maryland 1997

PATENT NUMBER: WO 9736998 PATENT DATE: 971009 WPI ACCESSION NO.: 97-503091 (9746)

PRIORITY APPLIC. NO.: US 624125 APPLIC. DATE: 960329

NATIONAL APPLIC. NO.: WO 97US540 APPLIC. DATE: 970128

LANGUAGE: English

ABSTRACT: Isolated eukaryotic enzymes having the 524 (Arabidopsis thaliana epsilon-cyclase), 294 (A. thaliana beta-**carotene** -hydroxylase), 305 or 293 (Haematococcus pluvialis isopentenyl-pyrophosphate (IPP) -isomerase) and 284 or 261 (A. thaliana IPP -isomerase) amino acid protein sequences (specified) are new. Also claimed are: an isolated DNA sequence, preferably having the 1,860, 954, 996, 1,165, 1,135 or 956 bp DNA sequence (specified), encoding the 294, 305, 293, 284, 261 or 294 residue sequences; an expression vector, preferably plasmid pATeps, plasmid pHP05, plasmid pMDP1, plasmid pATDP7, plasmid pHP04 or plasmid pAT0HB (ATCC 98005, 98000, 98001, 98002, 98004 or 98003), containing the DNA sequence; a host cell (e.g. Escherichia coli) containing the expression vector; and a DNA sequence which results in the expression of a eukaryotic **carotenoid** biosynthetic enzyme. The following methods are also claimed: screening for eukaryotic genes involved in **carotenoid** biosynthesis, metabolism or degradation; producing a **carotenoid**; inhibiting **carotenoid** synthesis in a host; increasing the production of a secondary metabolite of IPP -isomerase by a host. (89pp)

8/3,AB/24 (Item 2 from file: 357)
DIALOG(R)File 357:Derwent Biotechnology Abs
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214840 DBA Accession No.: 97-09961 PATENT
**Phaffia derived GAPDH and carotenoid synthesis genes and promoter
fragment- for carotenoid or therapeutic recombinant protein
production in Phaffia rhodozyma**
AUTHOR: Verdoes J C; Wery J
CORPORATE SOURCE: Delft, The Netherlands; Voorburg, The Netherlands.
PATENT ASSIGNEE: Brocades; Ooijen A J 1997
PATENT NUMBER: WO 9723633 PATENT DATE: 970703 WPI ACCESSION NO.:
97-351068 (9732)
PRIORITY APPLIC. NO.: EP 96200943 APPLIC. DATE: 960411
NATIONAL APPLIC. NO.: WO 96EP5887 APPLIC. DATE: 961223
LANGUAGE: English

ABSTRACT: A new DNA sequence contains a promoter with a region found upstream of the open reading frame of a highly expressed *Phaffia* sp. gene, e.g. from a glycolytic pathway (preferably encoding glyceraldehyde-3-phosphate-dehydrogenase, GAPDH, EC-1.2.1.12), or encoding a ribosome protein. A heterologous downstream sequence may be included, with an open reading frame encoding a protein responsible for reduced sensitivity to a selective agent, e.g. G418-resistance, or a carotenoid biosynthesis pathway enzyme, e.g. isopentenyl - pyrophosphate - isomerase, geranylgeranyl-pyrophosphate-synthase, phytoene-synthase, phytoene-desaturase or lycopene-cyclase. A GAPDH transcription terminator may be inserted downstream of the gene. The DNA may be inserted in a vector for expression in *Phaffia rhodozyma*, optionally with integration of at least 50 copies in the genome. A new method for isolation of a promoter involves cloning upstream sequences from highly-expressed genes from a *Phaffia* sp. cDNA library. The recombinant host may be used to produce carotenoids or therapeutic recombinant proteins (claimed). (118pp)

8/3,AB/25 (Item 3 from file: 357)
DIALOG(R)File 357:Derwent Biotechnology Abs
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006795 DBA Accession No.: 83-01991
**Biosynthesis of allylic isoprenoid pyrophosphates by an enzyme preparation
from the flavedo of Citrus paradisi- characterization of isopentenyl
pyrophosphate- isomerase and prenyltransferase from grapefruit
flavedo**
AUTHOR: Perez L M; Lozada R; Cori O
CORPORATE SOURCE: Laboratorio de Bioquimica General, Facultad de Ciencias
Basicas y Farmaceuticas, Universidad de Chile, Casilla 233, Santiago 1,
Chile.
JOURNAL: Phytochemistry (22, 2, 431-33) 1983
CODEN: PYTCAS
LANGUAGE: English

ABSTRACT: The flavedo of ripe grapefruits (*Citrus paradisi*) was homogenized and filtered. The cell-free extract transformed 3(RS)-(2-14C)mevalonic acid into mono- and sesquiterpenoids of E- and Z-conformation in 11.2% yield after 2 hr, in the presence of ATP, Mn²⁺ and sulfhydryl group protectors. (1-14C)Isopentenyl pyrophosphate (IPP) was transformed into 3,3'-dimethylallyl pyrophosphate. The activity of IPP -isomerase (EC-5.3.3.2) was 0.02 nkat/mg under initial rate conditions. The enzyme did not require the presence of Mg²⁺ or Mn²⁺, and was not inhibited by EDTA at concentrations up to 10 mM. Prenyltransferase (EC-2.5.1.1) had a pH optimum of 6.5. The activity of the enzyme was 0.038 nkat/mg at this pH value. Prenyltransferase was inactivated by 5,5'-dithio-bis(nitrobenzoic acid) and required bivalent metals for activity. The nature of the metal ion defined the stereochemistry of the products formed by prenyltransferase. Grapefruit could be a good starting material for the study of the stereochemistry

of the enzymes forming E and Z sesquiterpeneoids. (25 ref)

?ds

Set	Items	Description
S1	10553	IPP
S2	950	ISOPENT? (W) PYROPHOSPHATE?
S3	161	ISOPENT? (W) PYROPHOSPHATE? (W) ISOMERASE?
S4	88774	CAROTENE? OR CAROTENOID?
S5	16	S1 AND S3
S6	39	S3 AND S4
S7	52	S5 OR S6
S8	25	RD (unique items)
?		